

Claims

① (Amended) A method for pretreating a sample, which contains various lipoproteins, prior to measuring cholesterol existing in specific one of said lipoproteins in said sample, which comprises causing an enzyme, which acts upon free cholesterol as a substrate, to act upon said sample to consume only said free cholesterol in advance under conditions that said lipoproteins remain substantially unchanged.

2. (Amended) A method for pretreating a sample, which contains various lipoproteins, prior to measuring cholesterol existing in specific one of said lipoproteins in said sample, which comprises causing an enzyme, which acts upon free cholesterol as a substrate, and a reaction accelerator, which is selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, to act upon said sample to consume only said free cholesterol in advance under conditions that said lipoproteins remain substantially unchanged.

3. A pretreatment method according to claim 1 or 2, wherein said enzyme is cholesterol oxidase or cholesterol dehydrogenase.

④ (Amended) A method for quantitating cholesterol existing in a specific lipoprotein in a sample, which comprises causing an enzyme, which acts upon free cholesterol as a substrate,

to act upon said sample with said lipoprotein contained therein to consume only said free cholesterol under conditions that said lipoproteins remain substantially unchanged; and then measuring said cholesterol, which exists in said specific lipoprotein, by using a substance which acts upon said specific lipoprotein only.

5. (Amended) A method for quantitating cholesterol existing in a specific lipoprotein in a sample, which comprises causing an enzyme, which acts upon free cholesterol as a substrate, and a reaction accelerator, which is selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, to act upon said sample with said lipoprotein contained therein to consume only said free cholesterol and then measuring said cholesterol, which exists in said specific lipoprotein, by using a substance which acts upon said specific lipoprotein only.

6. A quantitation method according to claim 4 or 5, wherein said enzyme is cholesterol oxidase and/or cholesterol dehydrogenase.

7. A quantitation method according to any one of claims 4-6, wherein said specific lipoprotein is high density lipoprotein.

8. A pretreatment agent for a sample to be measured for

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cholesterol, comprising an enzyme, which acts upon free cholesterol as a substrate, and substantially no substance which acts upon lipoprotein.

9. A pretreatment agent for a sample to be measured for cholesterol, comprising an enzyme, which acts upon free cholesterol as a substrate, a reaction accelerator which is selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, and substantially no substance which acts upon lipoprotein.

10. A pretreatment agent for a sample to be measured for

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cholesterol, comprising an enzyme, which acts upon free cholesterol as a substrate, and substantially no cholesterol esterase.

11. A pretreatment agent for a sample to be measured for cholesterol, comprising an enzyme, which acts upon free cholesterol as a substrate, a reaction accelerator which is selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, and substantially no cholesterol esterase.

12. A pretreatment agent according to ~~any one of claims 8-10, wherein said enzyme is cholesterol dehydrogenase or cholesterol oxidase.~~

13. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising cholesterol oxidase and a hydrogen peroxide consuming substance; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only, cholesterol esterase, and a color developer.

14. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising cholesterol dehydrogenase and a coenzyme; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only, and cholesterol esterase.

15. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising cholesterol dehydrogenase and a coenzyme; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only, cholesterol oxidase, cholesterol esterase, peroxidase, and a color developer.

16. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising cholesterol oxidase, cholesterol esterase, and a hydrogen peroxide consuming substance; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only, and a color developer.

17. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising cholesterol dehydrogenase, a coenzyme, and cholesterol esterase; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only.

18. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising cholesterol dehydrogenase, a coenzyme, and cholesterol esterase; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only, cholesterol oxidase,

peroxidase, and a color developer.

19. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising cholesterol oxidase and a hydrogen peroxide consuming substance;

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only; and

(3) a third reagent comprising cholesterol esterase and a color developer.

20. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising cholesterol dehydrogenase and a coenzyme;

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only; and

(3) a third reagent comprising cholesterol esterase.

21. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising cholesterol dehydrogenase and a coenzyme;

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only; and

(3) a third reagent comprising cholesterol oxidase, cholesterol esterase, peroxidase, and a color developer.

22. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising cholesterol dehydrogenase, a coenzyme, and a coenzyme reaction product consuming substance; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only, and cholesterol esterase.

23. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising cholesterol dehydrogenase, a coenzyme, and a coenzyme reaction product consuming substance; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only, cholesterol esterase, and a color developer.

24. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising (a) cholesterol oxidase, (b) a reaction accelerator selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, and (c) a hydrogen peroxide consuming substance; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only, cholesterol esterase, and a color developer.

25. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising (a) cholesterol

dehydrogenase, (b) a reaction accelerator selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, and (c) a coenzyme; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only, and cholesterol esterase.

26. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising (a) cholesterol oxidase, (b) a reaction accelerator selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, (c) cholesterol esterase, and (d) a hydrogen peroxide consuming substance; and

(2) a second reagent comprising a substance, which acts upon said specific lipoprotein only, and a color developer.

27. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising (a) cholesterol dehydrogenase, (b) a coenzyme, (c) a reaction accelerator selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, and (d) cholesterol esterase; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only.

28. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising (a) cholesterol dehydrogenase, (b) a coenzyme, (c) a reaction accelerator selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, and (d) cholesterol esterase; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only, cholesterol oxidase, peroxidase, and a color developer.

29. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising (a) cholesterol oxidase, (b) a reaction accelerator selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, and (c) a hydrogen peroxide consuming substance;

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only; and

(3) a third reagent comprising cholesterol esterase and a color developer.

30. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising (a) cholesterol dehydrogenase, (b) a reaction accelerator selected from

flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, and (c) a coenzyme;

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only; and

(3) a third reagent comprising cholesterol esterase.

31. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising (a) cholesterol dehydrogenase, (b) a reaction accelerator selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, and (c) a coenzyme;

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only; and

(3) a third reagent comprising cholesterol oxidase, cholesterol esterase, peroxidase, and a color developer.

32. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising (a) cholesterol dehydrogenase, (b) a coenzyme, (c) a reaction accelerator selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, and (d) coenzyme reaction product consuming substance; and

(2) a second reagent comprising a substance which acts

upon said specific lipoprotein only, and cholesterol esterase.

33. A quantitation kit for cholesterol in a specific lipoprotein, comprising the following reagents:

(1) a first reagent comprising (a) cholesterol dehydrogenase, (b) a coenzyme, (c) a reaction accelerator selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, and (d) coenzyme reaction product consuming substance; and

(2) a second reagent comprising a substance which acts upon said specific lipoprotein only, cholesterol esterase, and a color developer.

34. A reaction accelerator for an enzyme capable of acting upon free cholesterol as a substrate, which is selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin.

35. A reaction accelerator according to ~~claim 34~~, wherein said enzyme is cholesterol oxidase or cholesterol dehydrogenase.

(36). A method for quantitating free cholesterol, which comprises causing an enzyme, which acts upon free cholesterol as a substrate, and a reaction accelerator, which is selected from flufenamic acid, mefenamic acid, 2,2',6',2"-terpyridine, tiglic acid, fusidic acid, betamethasone acetate, monensin or mevinolin, to act.

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